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UTILITY PATENT APPLICATION TRANSMITTAL <i>(Only for new nonprovisional applications under 37 C.F.R. § 1.53 (b))</i>		Attorney Docket No. LEX-0081-USA
First Inventor or Application Identifier Marie Harras et al.		Title Novel Human Transporter Proteins and Polynucleotide Encoding the Same
Express Mail label No. EL672756229US		

APPLICATION ELEMENTS <i>See MPEP chapter 600 concerning utility patent application contents</i>	ADDRESS TO: Assistant Commissioner for Patents Box Patent Application Washington, DC 20231
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<p>1. <input type="checkbox"/> *Fee Transmittal Form (e.g., PTO/SB/17) <i>(Submit an original and a duplicate for fee processing)</i></p> <p>2. <input checked="" type="checkbox"/> Specification [Total 25] <i>(preferred arrangement set forth below)</i></p> <ul style="list-style-type: none"> - Descriptive title of the invention - Cross References to Related Applications - Statement Regarding Fed sponsored R & D - Reference to Microfiche Appendix - Background of the Invention - Brief Summary of the Invention - Brief Description of the drawings (if filed) - Detailed Description - Claim(s) - Abstract of the disclosure <p>3. <input type="checkbox"/> Drawing(s) (35 U.S.C. 113) [Total 1] <i>Sheets</i></p> <p>4. Oath or Declaration [Total 1]</p> <p>a. <input checked="" type="checkbox"/> Newly unexecuted (original or copy)</p> <p>b. <input type="checkbox"/> Copy from a prior application (37 C.F.R. § 1.63(d)) <i>(for continuation/divisional with Box 16 completed)</i></p> <p>i. <input type="checkbox"/> DELETION OF INVENTOR(S) Signed statement attached deleting inventor(s) named in the prior application, see 37 C.F.R. §§ 1.63(d)(2) and 1.33(b).</p>	<p>5. <input type="checkbox"/> Microfiche Computer Program (Appendix)</p> <p>6. Nucleotide and/or Amino Acid Sequence Submission <i>(if applicable, all necessary)</i></p> <p>a. <input type="checkbox"/> Computer Readable Copy</p> <p>b. <input checked="" type="checkbox"/> Paper Copy (identical to computer copy)</p> <p>c. <input type="checkbox"/> Statement verifying identity of above copies</p>
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Signature		Date	October 31, 2000

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**STATEMENT CLAIMING SMALL ENTITY STATUS
(37 CFR 1.9(f) & 1.27(c)) -- SMALL BUSINESS CONCERN**

Docket Number (Optional)
LEX-0081-USA

Applicant, Patentee, or Identifier: Marie Harras et al.

Application or Patent No.: _____

Filed or Issued: October 31, 2000

Title: Novel Human Transporter Proteins and Polynucleotides Encoding the Same

I hereby state that I am

- ☐ the owner of the small business concern identified below:
☒ an official of the small business concern empowered to act on behalf of the concern identified below:

NAME OF SMALL BUSINESS CONCERN Lexicon Genetics Incorporated

ADDRESS OF SMALL BUSINESS CONCERN 4000 Research Forest Drive, The Woodlands, TX 77381

I hereby state that the above identified small business concern qualifies as a small business concern as defined in 13 CFR Part 121 for purposes of paying reduced fees to the United States Patent and Trademark Office. Questions related to size standards for a small business concern may be directed to: Small Business Administration, Size Standards Staff, 409 Third Street, SW, Washington, DC 20416.

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☐ the application identified above.
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If the rights held by the above identified small business concern are not exclusive, each individual, concern, or organization having rights in the invention must file separate statements as to their status as small entities, and no rights to the invention are held by any person, other than the inventor, who would not qualify as an independent inventor under 37 CFR 1.9(c) if that person made the invention, or by any concern which would not qualify as a small business concern under 37 CFR 1.9(d), or a nonprofit organization under 37 CFR 1.9(e).

Each person concern, or organization having any rights in the invention is listed below:

- ☒ No such persons, concerns, or organizations exist.
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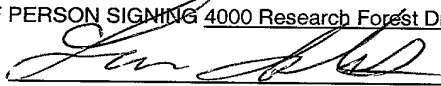
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NAME OF PERSON SIGNING Lance K. Ishimoto

TITLE OF PERSON IF OTHER THAN OWNER Vice President - Intellectual Property

ADDRESS OF PERSON SIGNING 4000 Research Forest Drive, The Woodlands, TX 77381

SIGNATURE 
Reg. No. 41,866

DATE October 31, 2000

**NOVEL HUMAN TRANSPORTER PROTEINS AND
POLYNUCLEOTIDES ENCODING THE SAME**

5 The present application claims the benefit of U.S.
Provisional Application Number 60/163,018 which was filed on
November 2, 1999 and is herein incorporated by reference in its
entirety.

1. INTRODUCTION

10 The present invention relates to the discovery,
identification, and characterization of novel human
polynucleotides encoding proteins that share sequence similarity
with mammalian transporter proteins. The invention encompasses
the described polynucleotides, host cell expression systems, the
15 encoded proteins, fusion proteins, polypeptides and peptides,
antibodies to the encoded proteins and peptides, and genetically
engineered animals that either lack or over express the disclosed
genes, antagonists and agonists of the proteins, and other
compounds that modulate the expression or activity of the proteins
20 encoded by the disclosed genes that can be used for diagnosis,
drug screening, clinical trial monitoring, the treatment of
diseases and disorders, or otherwise contributing to the quality
of life.

2. BACKGROUND OF THE INVENTION

25 Transporter proteins are integral membrane proteins that
mediate or facilitate the passage of materials across the lipid
bilayer. Given that the transport of materials across the
membrane can play an important physiological role, transporter
proteins are good drug targets. Additionally, one of the
30 mechanisms of drug resistance involves diseased cells using
cellular transporter systems to export chemotherapeutic agents
from the cell. Such mechanisms are particularly relevant to cells
manifesting resistance to a multiplicity of drugs.

3. SUMMARY OF THE INVENTION

The present invention relates to the discovery, identification, and characterization of nucleotides that encode novel human proteins, and the corresponding amino acid sequences of these proteins. The novel human proteins (NHPs) described for the first time herein share structural similarity with mammalian multi-drug resistance (MDR) proteins and cellular transporters.

The novel human nucleic acid sequences described herein, encode alternative proteins/open reading frames (ORFs) of 659, 705, 1,063, 496, 542, 900, 978, 1,024, 1,382, 815, 861, 1,219, and amino acids in length (see SEQ ID NOS: 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 22, and 24 respectively).

The invention also encompasses agonists and antagonists of the described NHPs, including small molecules, large molecules, mutant NHPs, or portions thereof that compete with native NHP, peptides, and antibodies, as well as nucleotide sequences that can be used to inhibit the expression of the described NHPs (e.g., antisense and ribozyme molecules, and gene or regulatory sequence replacement constructs) or to enhance the expression of the described NHP genes (e.g., expression constructs that place the described gene under the control of a strong promoter system), and transgenic animals that express a NHP transgene, or "knock-outs" (which can be conditional) that do not express a functional NHP. A knockout ES cell line has been produced that contains a gene trap mutation in the murine ortholog of the described gene.

Further, the present invention also relates to processes for identifying compounds that modulate, *i.e.*, act as agonists or antagonists, of NHP expression and/or NHP activity that utilize purified preparations of the described NHPs and/or NHP product, or cells expressing the same. Such compounds can be used as therapeutic agents for the treatment of any of a wide variety of symptoms associated with biological disorders or imbalances.

4. DESCRIPTION OF THE SEQUENCE LISTING AND FIGURES

The Sequence Listing provides the sequences of the described NHP ORFs that encode the described NHP amino acid sequences. SEQ ID NO:25 describes a NHP ORF as well as flanking 5' and 3'

5 sequences.

5. DETAILED DESCRIPTION OF THE INVENTION

The NHPs, described for the first time herein, are novel proteins that are expressed in, *inter alia*, human cell lines, predominantly in human mammary gland, as well as human fetal liver, prostate, testis, and gene trapped human cells.

The present invention encompasses the nucleotides presented in the Sequence Listing, host cells expressing such nucleotides, the expression products of such nucleotides, and: (a) nucleotides that encode mammalian homologs of the described genes, including the specifically described NHPs, and the NHP products; (b) nucleotides that encode one or more portions of the NHPs that correspond to functional domains, and the polypeptide products specified by such nucleotide sequences, including but not limited to the novel regions of any active domain(s); (c) isolated nucleotides that encode mutant versions, engineered or naturally occurring, of the described NHPs in which all or a part of at least one domain is deleted or altered, and the polypeptide products specified by such nucleotide sequences, including but not limited to soluble proteins and peptides in which all or a portion of the signal sequence is deleted; (d) nucleotides that encode chimeric fusion proteins containing all or a portion of a coding region of an NHP, or one of its domains (e.g., a receptor or ligand binding domain, accessory protein/self-association domain, etc.) fused to another peptide or polypeptide; or (e) therapeutic or diagnostic derivatives of the described polynucleotides such as oligonucleotides, antisense polynucleotides, ribozymes, dsRNA, or gene therapy constructs comprising a sequence first disclosed in

the Sequence Listing. As discussed above, the present invention includes: (a) the human DNA sequences presented in the Sequence Listing (and vectors comprising the same) and additionally contemplates any nucleotide sequence encoding a contiguous NHP open reading frame (ORF) that hybridizes to a complement of a DNA sequence presented in the Sequence Listing under highly stringent conditions, e.g., hybridization to filter-bound DNA in 0.5 M NaHPO₄, 7% sodium dodecyl sulfate (SDS), 1 mM EDTA at 65°C, and washing in 0.1xSSC/0.1% SDS at 68°C (Ausubel F.M. et al., eds., 1989, Current Protocols in Molecular Biology, Vol. I, Green Publishing Associates, Inc., and John Wiley & sons, Inc., New York, at p. 2.10.3) and encodes a functionally equivalent gene product. Additionally contemplated are any nucleotide sequences that hybridize to the complement of a DNA sequence that encodes and expresses an amino acid sequence presented in the Sequence Listing under moderately stringent conditions, e.g., washing in 0.2xSSC/0.1% SDS at 42°C (Ausubel et al., 1989, *supra*), yet still encodes a functionally equivalent NHP product. Functional equivalents of a NHP include naturally occurring NHPs present in other species and mutant NHPs whether naturally occurring or engineered (by site directed mutagenesis, gene shuffling, directed evolution as described in, for example, U.S. Patent No. 5,837,458). The invention also includes degenerate nucleic acid variants of the disclosed NHP polynucleotide sequences.

Additionally contemplated are polynucleotides encoding NHP ORFs, or their functional equivalents, encoded by polynucleotide sequences that are about 99, 95, 90, or about 85 percent similar or identical to corresponding regions of the nucleotide sequences of the Sequence Listing (as measured by BLAST sequence comparison analysis using, for example, the GCG sequence analysis package using standard default settings).

The invention also includes nucleic acid molecules, preferably DNA molecules, that hybridize to, and are therefore the complements of, the described NHP gene nucleotide sequences. Such hybridization conditions may be highly stringent or less highly stringent, as described above. In instances where the nucleic acid molecules are deoxyoligonucleotides ("DNA oligos"), such molecules are generally about 16 to about 100 bases long, or about 20 to about 80, or about 34 to about 45 bases long, or any variation or combination of sizes represented therein that incorporate a contiguous region of sequence first disclosed in the Sequence Listing. Such oligonucleotides can be used in conjunction with the polymerase chain reaction (PCR) to screen libraries, isolate clones, and prepare cloning and sequencing templates, etc..

Alternatively, such NHP oligonucleotides can be used as hybridization probes for screening libraries, and assessing gene expression patterns (particularly using a micro array or high-throughput "chip" format). Additionally, a series of the described NHP oligonucleotide sequences, or the complements thereof, can be used to represent all or a portion of the described NHP sequences. The oligonucleotides, typically between about 16 to about 40 (or any whole number within the stated range) nucleotides in length can partially overlap each other and/or the NHP sequence may be represented using oligonucleotides that do not overlap. Accordingly, the described NHP polynucleotide sequences shall typically comprise at least about two or three distinct oligonucleotide sequences of at least about 18, and preferably about 25, nucleotides in length that are each first disclosed in the described Sequence Listing. Such oligonucleotide sequences may begin at any nucleotide present within a sequence in the Sequence Listing and proceed in either a sense (5'-to-3') orientation vis-a-vis the described sequence or in an antisense orientation.

For oligonucleotide probes, highly stringent conditions may refer, e.g., to washing in 6xSSC/0.05% sodium pyrophosphate at 37°C (for 14-base oligos), 48°C (for 17-base oligos), 55°C (for 20-base oligos), and 60°C (for 23-base oligos). These nucleic acid molecules may encode or act as NHP gene antisense molecules, useful, for example, in NHP gene regulation (for and/or as antisense primers in amplification reactions of NHP gene nucleic acid sequences). With respect to NHP gene regulation, such techniques can be used to regulate biological functions. Further, such sequences may be used as part of ribozyme and/or triple helix sequences that are also useful for NHP gene regulation.

Inhibitory antisense or double stranded oligonucleotides can additionally comprise at least one modified base moiety which is selected from the group including but not limited to

- 5-fluorouracil, 5-bromouracil, 5-chlorouracil, 5-iodouracil, hypoxanthine, xantine, 4-acetylcytosine, 5-(carboxyhydroxymethyl)uracil, 5-carboxymethylaminomethyl-2-thiouridine, 5-carboxymethylaminomethyluracil, dihydrouracil, beta-D-galactosylqueosine, inosine, N6-isopentenyladenine, 1-methylguanine, 1-methylinosine, 2,2-dimethylguanine, 2-methyladenine, 2-methylguanine, 3-methylcytosine, 5-methylcytosine, N6-adenine, 7-methylguanine, 5-methylaminomethyluracil, 5-methoxyaminomethyl-2-thiouracil, beta-D-mannosylqueosine, 5'-methoxycarboxymethyluracil, 5-methoxyuracil, 2-methylthio-N6-isopentenyladenine, uracil-5-oxyacetic acid (v), wybutoxosine, pseudouracil, queosine, 2-thiocytosine, 5-methyl-2-thiouracil, 2-thiouracil, 4-thiouracil, 5-methyluracil, uracil-5-oxyacetic acid methylester, uracil-5-oxyacetic acid (v), 5-methyl-2-thiouracil, 3-(3-amino-3-N-2-carboxypropyl) uracil, (acp3)w, and 2,6-diaminopurine.

The antisense oligonucleotide can also comprise at least one modified sugar moiety selected from the group including but not limited to arabinose, 2-fluoroarabinose, xylulose, and hexose.

In yet another embodiment, the antisense oligonucleotide will comprise at least one modified phosphate backbone selected from the group consisting of a phosphorothioate, a phosphorodithioate, a phosphoramidothioate, a phosphoramidate, a phosphordiamidate, a methylphosphonate, an alkyl phosphotriester, and a formacetal or analog thereof.

In yet another embodiment, the antisense oligonucleotide is an α -anomeric oligonucleotide. An α -anomeric oligonucleotide forms specific double-stranded hybrids with complementary RNA in which, contrary to the usual β -units, the strands run parallel to each other (Gautier *et al.*, 1987, Nucl. Acids Res. 15:6625-6641). The oligonucleotide is a 2'-O-methylribonucleotide (Inoue *et al.*, 1987, Nucl. Acids Res. 15:6131-6148), or a chimeric RNA-DNA analogue (Inoue *et al.*, 1987, FEBS Lett. 215:327-330). Alternatively, double stranded RNA can be used to disrupt the expression and function of a targeted NHP.

Oligonucleotides of the invention can be synthesized by standard methods known in the art, e.g. by use of an automated DNA synthesizer (such as are commercially available from Biosearch, Applied Biosystems, etc.). As examples, phosphorothioate oligonucleotides can be synthesized by the method of Stein *et al.* (1988, Nucl. Acids Res. 16:3209), and methylphosphonate oligonucleotides can be prepared by use of controlled pore glass polymer supports (Sarin *et al.*, 1988, Proc. Natl. Acad. Sci. U.S.A. 85:7448-7451), etc.

Low stringency conditions are well known to those of skill in the art, and will vary predictably depending on the specific organisms from which the library and the labeled sequences are derived. For guidance regarding such conditions see, for example, Sambrook *et al.*, 1989, Molecular Cloning, A Laboratory Manual (and periodic updates thereof), Cold Springs Harbor Press, N.Y.; and

Ausubel et al., 1989, Current Protocols in Molecular Biology, Green Publishing Associates and Wiley Interscience, N.Y.

Alternatively, suitably labeled NHP nucleotide probes can be used to screen a human genomic library using appropriately
5 stringent conditions or by PCR. The identification and characterization of human genomic clones is helpful for identifying polymorphisms (including, but not limited to, nucleotide repeats, microsatellite alleles, single nucleotide polymorphisms, or coding single nucleotide polymorphisms),
10 determining the genomic structure of a given locus/allele, and designing diagnostic tests. For example, sequences derived from regions adjacent to the intron/exon boundaries of the human gene can be used to design primers for use in amplification assays to detect mutations within the exons, introns, splice sites (e.g.,
15 splice acceptor and/or donor sites), etc., that can be used in diagnostics and pharmacogenomics.

Further, a NHP gene homolog can be isolated from nucleic acid from an organism of interest by performing PCR using two degenerate or "wobble" oligonucleotide primer pools designed on
20 the basis of amino acid sequences within the NHP products disclosed herein. The template for the reaction may be total RNA, mRNA, and/or cDNA obtained by reverse transcription of mRNA prepared from human or non-human cell lines or tissue known or suspected to express an allele of a NHP gene.

25 The PCR product can be subcloned and sequenced to ensure that the amplified sequences represent the sequence of the desired NHP gene. The PCR fragment can then be used to isolate a full length cDNA clone by a variety of methods. For example, the amplified fragment can be labeled and used to screen a cDNA library, such as
30 a bacteriophage cDNA library. Alternatively, the labeled fragment can be used to isolate genomic clones via the screening of a genomic library.

PCR technology can also be used to isolate full length cDNA sequences. For example, RNA can be isolated, following standard procedures, from an appropriate cellular or tissue source (*i.e.*, one known, or suspected, to express a NHP gene). A reverse transcription (RT) reaction can be performed on the RNA using an oligonucleotide primer specific for the most 5' end of the amplified fragment for the priming of first strand synthesis. The resulting RNA/DNA hybrid may then be "tailed" using a standard terminal transferase reaction, the hybrid may be digested with RNase H, and second strand synthesis may then be primed with a complementary primer. Thus, cDNA sequences upstream of the amplified fragment can be isolated. For a review of cloning strategies that can be used, see *e.g.*, Sambrook *et al.*, 1989, *supra*.

A cDNA encoding a mutant NHP gene can be isolated, for example, by using PCR. In this case, the first cDNA strand may be synthesized by hybridizing an oligo-dT oligonucleotide to mRNA isolated from tissue known or suspected to be expressed in an individual putatively carrying a mutant NHP allele, and by extending the new strand with reverse transcriptase. The second strand of the cDNA is then synthesized using an oligonucleotide that hybridizes specifically to the 5' end of the normal gene. Using these two primers, the product is then amplified via PCR, optionally cloned into a suitable vector, and subjected to DNA sequence analysis through methods well known to those of skill in the art. By comparing the DNA sequence of the mutant NHP allele to that of a corresponding normal NHP allele, the mutation(s) responsible for the loss or alteration of function of the mutant NHP gene product can be ascertained.

Alternatively, a genomic library can be constructed using DNA obtained from an individual suspected of or known to carry a mutant NHP allele (*e.g.*, a person manifesting a NHP-associated phenotype such as, for example, obesity, high blood pressure,

connective tissue disorders, infertility, etc.), or a cDNA library can be constructed using RNA from a tissue known, or suspected, to express a mutant NHP allele. A normal NHP gene, or any suitable fragment thereof, can then be labeled and used as a probe to

5 identify the corresponding mutant NHP allele in such libraries. Clones containing mutant NHP gene sequences can then be purified and subjected to sequence analysis according to methods well known to those skilled in the art.

Additionally, an expression library can be constructed
10 utilizing cDNA synthesized from, for example, RNA isolated from a tissue known, or suspected, to express a mutant NHP allele in an individual suspected of or known to carry such a mutant allele. In this manner, gene products made by the putatively mutant tissue can be expressed and screened using standard antibody screening
15 techniques in conjunction with antibodies raised against a normal NHP product, as described below. (For screening techniques, see, for example, Harlow, E. and Lane, eds., 1988, "Antibodies: A Laboratory Manual", Cold Spring Harbor Press, Cold Spring Harbor.)

Additionally, screening can be accomplished by screening with
20 labeled NHP fusion proteins, such as, for example, alkaline phosphatase-NHP or NHP-alkaline phosphatase fusion proteins. In cases where a NHP mutation results in an expressed gene product with altered function (e.g., as a result of a missense or a frameshift mutation), polyclonal antibodies to a NHP are likely to
25 cross-react with a corresponding mutant NHP gene product. Library clones detected via their reaction with such labeled antibodies can be purified and subjected to sequence analysis according to methods well known in the art.

The invention also encompasses (a) DNA vectors that contain
30 any of the foregoing NHP coding sequences and/or their complements (*i.e.*, antisense); (b) DNA expression vectors that contain any of the foregoing NHP coding sequences operatively associated with a regulatory element that directs the expression of the coding

sequences (for example, baculo virus as described in U.S. Patent No. 5,869,336 herein incorporated by reference); (c) genetically engineered host cells that contain any of the foregoing NHP coding sequences operatively associated with a regulatory element that
5 directs the expression of the coding sequences in the host cell; and (d) genetically engineered host cells that express an endogenous NHP gene under the control of an exogenously introduced regulatory element (*i.e.*, gene activation). As used herein, regulatory elements include, but are not limited to,
10 inducible and non-inducible promoters, enhancers, operators and other elements known to those skilled in the art that drive and regulate expression. Such regulatory elements include but are not limited to the human cytomegalovirus (hCMV) immediate early gene, regulatable, viral elements
15 (particularly retroviral LTR promoters), the early or late promoters of SV40 adenovirus, the *lac* system, the *trp* system, the *TAC* system, the *TRC* system, the major operator and promoter regions of phage lambda, the control regions of fd coat protein, the promoter for 3-phosphoglycerate kinase
20 (PGK), the promoters of acid phosphatase, and the promoters of the yeast α -mating factors.

The present invention also encompasses antibodies and anti-idiotypic antibodies (including Fab fragments), antagonists and agonists of the NHP, as well as compounds or nucleotide constructs
25 that inhibit expression of a NHP gene (transcription factor inhibitors, antisense and ribozyme molecules, or gene or regulatory sequence replacement constructs), or promote the expression of a NHP (*e.g.*, expression constructs in which NHP coding sequences are operatively associated with expression
30 control elements such as promoters, promoter/enhancers, etc.).

The NHPs or NHP peptides, NHP fusion proteins, NHP nucleotide sequences, antibodies, antagonists and agonists can be useful for the detection of mutant NHPs or inappropriately expressed NHPs for

the diagnosis of disease. The NHP proteins or peptides, NHP fusion proteins, NHP nucleotide sequences, host cell expression systems, antibodies, antagonists, agonists and genetically engineered cells and animals can be used for screening for drugs
5 (or high throughput screening of combinatorial libraries) effective in the treatment of the symptomatic or phenotypic manifestations of perturbing the normal function of NHP in the body. The use of engineered host cells and/or animals may offer an advantage in that such systems allow not only for the
10 identification of compounds that bind to the endogenous receptor for an NHP, but can also identify compounds that trigger NHP-mediated activities or pathways.

Finally, the NHP products can be used as therapeutics. For example, soluble derivatives such as NHP peptides/domains
15 corresponding the NHPs, NHP fusion protein products (especially NHP-Ig fusion proteins, *i.e.*, fusions of a NHP, or a domain of a NHP, to an IgFc), NHP antibodies and anti-idiotypic antibodies (including Fab fragments), antagonists or agonists (including compounds that modulate or act on downstream targets in a NHP-
20 mediated pathway) can be used to directly treat diseases or disorders. For instance, the administration of an effective amount of soluble NHP, or a NHP-IgFc fusion protein or an anti-idiotypic antibody (or its Fab) that mimics the NHP could activate or effectively antagonize the endogenous NHP receptor. Nucleotide
25 constructs encoding such NHP products can be used to genetically engineer host cells to express such products *in vivo*; these genetically engineered cells function as "bioreactors" in the body delivering a continuous supply of a NHP, a NHP peptide, or a NHP fusion protein to the body. Nucleotide constructs encoding
30 functional NHPs, mutant NHPs, as well as antisense and ribozyme molecules can also be used in "gene therapy" approaches for the modulation of NHP expression. Thus, the invention also

encompasses pharmaceutical formulations and methods for treating biological disorders.

Various aspects of the invention are described in greater detail in the subsections below.

5

5.1 THE NHP SEQUENCES

The cDNA sequences and the corresponding deduced amino acid sequences of the described NHPs are presented in the Sequence Listing. The NHP nucleotides were obtained from clustered human
10 gene trapped sequences, ESTs, and human testis and mammary gland cDNA libraries (Edge Biosystems, Gaithersburg, MD). The described sequences can also incorporate several coding region single nucleotide polymorphisms (cSNPs). The first polymorphism is a A to G transition at position 704 of, for example, SED ID NO: 23,
15 which results in a corresponding change of gln to an arg at, for example, position 235 of SEQ ID NO:24; the second can occur at position 2184 of, for example, SEQ ID NO:23 that changes a gln to a his at position 728 of, for example, SEQ ID NO:24; and the third cSNP involves a silent T to C transition at position 2,436 of, for
20 example, SEQ ID NO:23.

Similar MDR encoding sequences, uses, and applications that are germane to the described NHPs, are described in U.S. Patents Nos. 5,198,344 and 5,866,699 which are herein incorporated by reference in their entirety.

25

5.2 NHPS AND NHP POLYPEPTIDES

NHPs, polypeptides, peptide fragments, mutated, truncated, or deleted forms of the NHPs, and/or NHP fusion proteins can be prepared for a variety of uses. These uses include but are not
30 limited to the generation of antibodies, as reagents in diagnostic assays, for the identification of other cellular gene products related to a NHP, as reagents in assays for screening for compounds that can be as pharmaceutical reagents useful in the

therapeutic treatment of mental, biological, or medical disorders and diseases. Given the similarity information and expression data, the described NHPs can be targeted (by drugs, oligos, antibodies, etc,) in order to treat disease, or to therapeutically
5 augment the efficacy of, for example, chemotherapeutic agents used in the treatment of breast or prostate cancer.

The Sequence Listing discloses the amino acid sequences encoded by the described NHP genes. The NHPs typically display have initiator methionines in DNA sequence contexts consistent
10 with a translation initiation site.

The NHP amino acid sequences of the invention include the amino acid sequence presented in the Sequence Listing as well as analogues and derivatives thereof. Further, corresponding NHP homologues from other species are encompassed by the invention.
15 In fact, any NHP protein encoded by the NHP nucleotide sequences described above are within the scope of the invention, as are any novel polynucleotide sequences encoding all or any novel portion of an amino acid sequence presented in the Sequence Listing. The degenerate nature of the genetic code is well known, and,
20 accordingly, each amino acid presented in the Sequence Listing, is generically representative of the well known nucleic acid "triplet" codon, or in many cases codons, that can encode the amino acid. As such, as contemplated herein, the amino acid sequences presented in the Sequence Listing, when taken together
25 with the genetic code (see, for example, Table 4-1 at page 109 of "Molecular Cell Biology", 1986, J. Darnell et al. eds., Scientific American Books, New York, NY, herein incorporated by reference) are generically representative of all the various permutations and combinations of nucleic acid sequences that can encode such amino
30 acid sequences.

The invention also encompasses proteins that are functionally equivalent to the NHPs encoded by the presently described nucleotide sequences as judged by any of a number of criteria,

including, but not limited to, the ability to bind and cleave a substrate of a NHP, or the ability to effect an identical or complementary downstream pathway, or a change in cellular metabolism (e.g., proteolytic activity, ion flux, tyrosine

5 phosphorylation, etc.). Such functionally equivalent NHP proteins include, but are not limited to, additions or substitutions of amino acid residues within the amino acid sequence encoded by the NHP nucleotide sequences described above, but which result in a silent change, thus producing a functionally equivalent gene
10 product. Amino acid substitutions may be made on the basis of similarity in polarity, charge, solubility, hydrophobicity, hydrophilicity, and/or the amphipathic nature of the residues involved. For example, nonpolar (hydrophobic) amino acids include alanine, leucine, isoleucine, valine, proline, phenylalanine,
15 tryptophan, and methionine; polar neutral amino acids include glycine, serine, threonine, cysteine, tyrosine, asparagine, and glutamine; positively charged (basic) amino acids include arginine, lysine, and histidine; and negatively charged (acidic) amino acids include aspartic acid and glutamic acid.

20 A variety of host-expression vector systems can be used to express the NHP nucleotide sequences of the invention. Where, as in the present instance, the NHP peptide or polypeptide is thought to be membrane protein, the hydrophobic regions of the protein can be excised and the resulting soluble peptide or polypeptide can be
25 recovered from the culture media. Such expression systems also encompass engineered host cells that express a NHP, or functional equivalent, *in situ*. Purification or enrichment of a NHP from such expression systems can be accomplished using appropriate detergents and lipid micelles and methods well known to those
30 skilled in the art. However, such engineered host cells themselves may be used in situations where it is important not only to retain the structural and functional characteristics of

the NHP, but to assess biological activity, e.g., in drug screening assays.

The expression systems that may be used for purposes of the invention include but are not limited to microorganisms such as
5 bacteria (e.g., *E. coli*, *B. subtilis*) transformed with recombinant bacteriophage DNA, plasmid DNA or cosmid DNA expression vectors containing NHP nucleotide sequences; yeast (e.g., *Saccharomyces*, *Pichia*) transformed with recombinant yeast expression vectors containing NHP nucleotide sequences; insect cell systems infected
10 with recombinant virus expression vectors (e.g., baculovirus) containing NHP sequences; plant cell systems infected with recombinant virus expression vectors (e.g., cauliflower mosaic virus, CaMV; tobacco mosaic virus, TMV) or transformed with recombinant plasmid expression vectors (e.g., Ti plasmid)
15 containing NHP nucleotide sequences; or mammalian cell systems (e.g., COS, CHO, BHK, 293, 3T3) harboring recombinant expression constructs containing promoters derived from the genome of mammalian cells (e.g., metallothionein promoter) or from mammalian viruses (e.g., the adenovirus late promoter; the vaccinia virus
20 7.5K promoter).

In bacterial systems, a number of expression vectors may be advantageously selected depending upon the use intended for the NHP product being expressed. For example, when a large quantity of such a protein is to be produced for the generation of
25 pharmaceutical compositions of or containing NHP, or for raising antibodies to a NHP, vectors that direct the expression of high levels of fusion protein products that are readily purified may be desirable. Such vectors include, but are not limited, to the *E. coli* expression vector pUR278 (Ruther et al., 1983, EMBO J.
30 2:1791), in which a NHP coding sequence may be ligated individually into the vector in frame with the *lacZ* coding region so that a fusion protein is produced; pIN vectors (Inouye &

Inouye, 1985, Nucleic Acids Res. 13:3101-3109; Van Heeke & Schuster, 1989, J. Biol. Chem. 264:5503-5509); and the like. pGEX vectors (Pharmacia or American Type Culture Collection) can also be used to express foreign polypeptides as fusion proteins with glutathione S-transferase (GST). In general, such fusion proteins are soluble and can easily be purified from lysed cells by adsorption to glutathione-agarose beads followed by elution in the presence of free glutathione. The PGEX vectors are designed to include thrombin or factor Xa protease cleavage sites so that the cloned target gene product can be released from the GST moiety.

In an insect system, *Autographa californica* nuclear polyhydrosis virus (AcNPV) is used as a vector to express foreign genes. The virus grows in *Spodoptera frugiperda* cells. A NHP gene coding sequence may be cloned individually into non-essential regions (for example the polyhedrin gene) of the virus and placed under control of an AcNPV promoter (for example the polyhedrin promoter). Successful insertion of NHP gene coding sequence will result in inactivation of the polyhedrin gene and production of non-occluded recombinant virus (i.e., virus lacking the proteinaceous coat coded for by the polyhedrin gene). These recombinant viruses are then used to infect *Spodoptera frugiperda* cells in which the inserted gene is expressed (e.g., see Smith et al., 1983, J. Virol. 46:584; Smith, U.S. Patent No. 4,215,051).

In mammalian host cells, a number of viral-based expression systems may be utilized. In cases where an adenovirus is used as an expression vector, the NHP nucleotide sequence of interest may be ligated to an adenovirus transcription/translation control complex, e.g., the late promoter and tripartite leader sequence. This chimeric gene may then be inserted in the adenovirus genome by in vitro or in vivo recombination. Insertion in a non-essential region of the viral genome (e.g., region E1 or E3) will result in a recombinant virus that is viable and capable of expressing a NHP product in infected hosts (e.g., See Logan &

Shenk, 1984, Proc. Natl. Acad. Sci. USA 81:3655-3659). Specific initiation signals may also be required for efficient translation of inserted NHP nucleotide sequences. These signals include the ATG initiation codon and adjacent sequences. In cases where an entire NHP gene or cDNA, including its own initiation codon and adjacent sequences, is inserted into the appropriate expression vector, no additional translational control signals may be needed. However, in cases where only a portion of a NHP coding sequence is inserted, exogenous translational control signals, including, perhaps, the ATG initiation codon, must be provided. Furthermore, the initiation codon must be in phase with the reading frame of the desired coding sequence to ensure translation of the entire insert. These exogenous translational control signals and initiation codons can be of a variety of origins, both natural and synthetic. The efficiency of expression may be enhanced by the inclusion of appropriate transcription enhancer elements, transcription terminators, etc. (See Bittner *et al.*, 1987, Methods in Enzymol. 153:516-544).

In addition, a host cell strain may be chosen that modulates the expression of the inserted sequences, or modifies and processes the gene product in the specific fashion desired. Such modifications (e.g., glycosylation) and processing (e.g., cleavage) of protein products may be important for the function of the protein. Different host cells have characteristic and specific mechanisms for the post-translational processing and modification of proteins and gene products. Appropriate cell lines or host systems can be chosen to ensure the correct modification and processing of the foreign protein expressed. To this end, eukaryotic host cells which possess the cellular machinery for proper processing of the primary transcript, glycosylation, and phosphorylation of the gene product may be used. Such mammalian host cells include, but are not limited to,

CHO, VERO, BHK, HeLa, COS, MDCK, 293, 3T3, WI38, and in particular, human cell lines.

For long-term, high-yield production of recombinant proteins, stable expression is preferred. For example, cell lines which stably express the NHP sequences described above can be engineered. Rather than using expression vectors which contain viral origins of replication, host cells can be transformed with DNA controlled by appropriate expression control elements (e.g., promoter, enhancer sequences, transcription terminators, polyadenylation sites, etc.), and a selectable marker. Following the introduction of the foreign DNA, engineered cells may be allowed to grow for 1-2 days in an enriched media, and then are switched to a selective media. The selectable marker in the recombinant plasmid confers resistance to the selection and allows cells to stably integrate the plasmid into their chromosomes and grow to form foci which in turn can be cloned and expanded into cell lines. This method may advantageously be used to engineer cell lines which express the NHP product. Such engineered cell lines may be particularly useful in screening and evaluation of compounds that affect the endogenous activity of the NHP product.

A number of selection systems may be used, including but not limited to the herpes simplex virus thymidine kinase (Wigler, et al., 1977, Cell 11:223), hypoxanthine-guanine phosphoribosyltransferase (Szybalska & Szybalski, 1962, Proc. Natl. Acad. Sci. USA 48:2026), and adenine phosphoribosyltransferase (Lowy, et al., 1980, Cell 22:817) genes can be employed in tk⁻, hgp⁻ or ap⁻ cells, respectively. Also, antimetabolite resistance can be used as the basis of selection for the following genes: dhfr, which confers resistance to methotrexate (Wigler, et al., 1980, Natl. Acad. Sci. USA 77:3567; O'Hare, et al., 1981, Proc. Natl. Acad. Sci. USA 78:1527); gpt, which confers resistance to mycophenolic acid (Mulligan & Berg, 1981, Proc. Natl. Acad. Sci. USA 78:2072); neo, which confers

resistance to the aminoglycoside G-418 (Colberre-Garapin, *et al.*, 1981, J. Mol. Biol. 150:1); and hygromycin (Santerre, *et al.*, 1984, Gene 30:147).

Alternatively, any fusion protein can be readily purified by
5 utilizing an antibody specific for the fusion protein being
expressed. For example, a system described by Janknecht *et al.*
allows for the ready purification of non-denatured fusion proteins
expressed in human cell lines (Janknecht, *et al.*, 1991, Proc.
Natl. Acad. Sci. USA 88:8972-8976). In this system, the gene of
10 interest is subcloned into a vaccinia recombination plasmid such
that the gene's open reading frame is translationally fused to an
amino-terminal tag consisting of six histidine residues. Extracts
from cells infected with recombinant vaccinia virus are loaded
onto Ni²⁺-nitriloacetic acid-agarose columns and histidine-tagged
15 proteins are selectively eluted with imidazole-containing buffers.

5.3 ANTIBODIES TO NHP PRODUCTS

Antibodies that specifically recognize one or more epitopes
of a NHP, or epitopes of conserved variants of a NHP, or peptide
20 fragments of a NHP are also encompassed by the invention. Such
antibodies include but are not limited to polyclonal antibodies,
monoclonal antibodies (mAbs), humanized or chimeric antibodies,
single chain antibodies, Fab fragments, F(ab')₂ fragments,
fragments produced by a Fab expression library, anti-idiotypic
25 (anti-Id) antibodies, and epitope-binding fragments of any of the
above.

The antibodies of the invention may be used, for example, in
the detection of NHP in a biological sample and may, therefore, be
utilized as part of a diagnostic or prognostic technique whereby
30 patients may be tested for abnormal amounts of NHP. Such
antibodies may also be utilized in conjunction with, for example,
compound screening schemes for the evaluation of the effect of
test compounds on expression and/or activity of a NHP gene

product. Additionally, such antibodies can be used in conjunction
gene therapy to, for example, evaluate the normal and/or
engineered NHP-expressing cells prior to their introduction into
the patient. Such antibodies may additionally be used as a method
5 for the inhibition of abnormal NHP activity. Thus, such
antibodies may, therefore, be utilized as part of treatment
methods.

For the production of antibodies, various host animals may be
immunized by injection with the NHP, an NHP peptide (e.g., one
10 corresponding the a functional domain of an NHP), truncated NHP
polypeptides (NHP in which one or more domains have been deleted),
functional equivalents of the NHP or mutated variant of the NHP.
Such host animals may include but are not limited to pigs,
rabbits, mice, goats, and rats, to name but a few. Various
15 adjuvants may be used to increase the immunological
response, depending on the host species, including but not
limited to Freund's adjuvant (complete and incomplete),
mineral salts such as aluminum hydroxide or aluminum
phosphate, surface active substances such as lysolecithin,
20 pluronic polyols, polyanions, peptides, oil emulsions, and
potentially useful human adjuvants such as BCG (bacille
Calmette-Guerin) and *Corynebacterium parvum*. Alternatively,
the immune response could be enhanced by combination and or
coupling with molecules such as keyhole limpet hemocyanin,
25 tetanus toxoid, diptheria toxoid, ovalbumin, cholera toxin
or fragments thereof. Polyclonal antibodies are
heterogeneous populations of antibody molecules derived from
the sera of the immunized animals.

Monoclonal antibodies, which are homogeneous populations of
30 antibodies to a particular antigen, can be obtained by any
technique which provides for the production of antibody molecules
by continuous cell lines in culture. These include, but are not
limited to, the hybridoma technique of Kohler and Milstein, (1975,

Nature 256:495-497; and U.S. Patent No. 4,376,110), the human B-cell hybridoma technique (Kosbor et al., 1983, Immunology Today 4:72; Cole et al., 1983, Proc. Natl. Acad. Sci. USA 80:2026-2030), and the EBV-hybridoma technique (Cole et al., 1985, Monoclonal
5 Antibodies And Cancer Therapy, Alan R. Liss, Inc., pp. 77-96). Such antibodies may be of any immunoglobulin class including IgG, IgM, IgE, IgA, IgD and any subclass thereof. The hybridoma producing the mAb of this invention may be cultivated in vitro or in vivo. Production of high titers of mAbs in vivo makes this the
10 presently preferred method of production.

In addition, techniques developed for the production of "chimeric antibodies" (Morrison et al., 1984, Proc. Natl. Acad. Sci., 81:6851-6855; Neuberger et al., 1984, Nature, 312:604-608; Takeda et al., 1985, Nature, 314:452-454) by splicing the genes
15 from a mouse antibody molecule of appropriate antigen specificity together with genes from a human antibody molecule of appropriate biological activity can be used. A chimeric antibody is a molecule in which different portions are derived from different animal species, such as those having a variable region derived
20 from a murine mAb and a human immunoglobulin constant region. Such technologies are described in U.S. Patents Nos. 6,075,181 and 5,877,397 and their respective disclosures which are herein incorporated by reference in their entirety.

25 Alternatively, techniques described for the production of single chain antibodies (U.S. Patent 4,946,778; Bird, 1988, Science 242:423-426; Huston et al., 1988, Proc. Natl. Acad. Sci. USA 85:5879-5883; and Ward et al., 1989, Nature 334:544-546) can be adapted to produce single chain antibodies against NHP gene
30 products. Single chain antibodies are formed by linking the heavy and light chain fragments of the Fv region via an amino acid bridge, resulting in a single chain polypeptide.

Antibody fragments which recognize specific epitopes may be generated by known techniques. For example, such fragments include, but are not limited to: the F(ab')₂ fragments which can be produced by pepsin digestion of the antibody molecule and the
5 Fab fragments which can be generated by reducing the disulfide bridges of the F(ab')₂ fragments. Alternatively, Fab expression libraries may be constructed (Huse et al., 1989, Science, 246:1275-1281) to allow rapid and easy identification of monoclonal Fab fragments with the desired specificity.

10 Antibodies to a NHP can, in turn, be utilized to generate anti-idiotypic antibodies that "mimic" a given NHP, using techniques well known to those skilled in the art. (See, e.g., Greenspan & Bona, 1993, FASEB J 7(5):437-444; and Nissinoff, 1991, J. Immunol. 147(8):2429-2438). For example antibodies which bind
15 to a NHP domain and competitively inhibit the binding of NHP to its cognate receptor can be used to generate anti-idiotypes that "mimic" the NHP and, therefore, bind and activate or neutralize a receptor. Such anti-idiotypic antibodies or Fab fragments of such anti-idiotypes can be used in therapeutic regimens involving a NHP
20 mediated pathway.

The present invention is not to be limited in scope by the specific embodiments described herein, which are intended as single illustrations of individual aspects of the invention, and functionally equivalent methods and components are within the
25 scope of the invention. Indeed, various modifications of the invention, in addition to those shown and described herein will become apparent to those skilled in the art from the foregoing description. Such modifications are intended to fall within the scope of the appended claims. All cited publications, patents,
30 and patent applications are herein incorporated by reference in their entirety.

WHAT IS CLAIMED IS:

1. An isolated nucleic acid molecule comprising at least 24 contiguous bases of nucleotide sequence first disclosed
5 in the NHP gene described in SEQ ID NO: 23.

2. An isolated nucleic acid molecule comprising a nucleotide sequence that:

- 10 (a) encodes the amino acid sequence shown in SEQ ID NO: 24; and
(b) hybridizes under stringent conditions to the nucleotide sequence of SEQ ID NO: 23 or the complement thereof.

15 3. An isolated nucleic acid molecule comprising a nucleotide sequence that:

- (a) encodes the amino acid sequence shown in SEQ ID NO: 2; and
20 (b) hybridizes under stringent conditions to the nucleotide sequence of SEQ ID NO: 1 or the complement thereof.

4. An isolated nucleic acid molecule comprising a nucleotide sequence that:

- 25 (a) encodes the amino acid sequence shown in SEQ ID NO: 48; and
(b) hybridizes under stringent conditions to the nucleotide sequence of SEQ ID NO: 47 or the
30 complement thereof.

ABSTRACT

Novel human polynucleotide and polypeptide sequences are disclosed that can be used in therapeutic, diagnostic, and pharmacogenomic applications.

PATENT APPLICATION

DECLARATION AND POWER OF ATTORNEY
FOR PATENT APPLICATION

ATTORNEY DOCKET NO. LEX-0081-USA

As a below named inventor, I hereby declare that:

My residence/post office address and citizenship are as stated below next to my name;

I believe I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled:

Novel Human Transporter Proteins and Polynucleotides Encoding the Same

the specification of which is attached hereto unless the following box is checked:

☐ was filed on _____ as US Application Serial No. or PCT International Application
Number _____ and was amended on _____ (if applicable).

I hereby state that I have reviewed and understood the contents of the above-identified specification, including the claims, as amended by any amendment(s) referred to above. I acknowledge the duty to disclose all information which is material to patentability as defined in 37 CFR 1.56.

Foreign Application(s) and/or Claim of Foreign Priority

I hereby claim foreign priority benefits under Title 35, United States Code Section 119 of any foreign application(s) for patent or inventor(s) certificate listed below and have also identified below any foreign application for patent or inventor(s) certificate having a filing date before that of the application on which priority is claimed:

COUNTRY	APPLICATION NUMBER	DATE FILED	PRIORITY CLAIMED UNDER 35 U.S.C. 119
			YES: _____ NO: _____
			YES: _____ NO: _____

Provisional Application

I hereby claim the benefit under Title 35, United States Code Section 119(e) of any United States provisional application(s) listed below:

APPLICATION SERIAL NUMBER	FILING DATE
60/163,018	11/2/1999

U.S. Priority Claim

I hereby claim the benefit under Title 35, United States Code, Section 120 of any United States application(s) listed below and, insofar as the subject matter of each of the claims of this application is not disclosed in the prior United States application in the manner provided by the first paragraph of Title 35, United States Code Section 112, I acknowledge the duty to disclose material information as defined in Title 37, Code of Federal Regulations, Section 1.56(a) which occurred between the filing date of the prior application and the national or PCT international filing date of this application:

APPLICATION SERIAL NUMBER	FILING DATE	STATUS(patented/pending/abandoned)

POWER OF ATTORNEY:

As a named inventor, I hereby appoint the following attorney(s) and/or agent(s) listed below to prosecute this application and transact all business in the Patent and Trademark Office connected therewith.

Lance K. Ishimoto, Reg. No. 41866

Send Correspondence to: Lance K. Ishimoto Lexicon Genetics Incorporated 4000 Research Forest Drive The Woodlands, TX 77381	Direct Telephone Calls To: Lance K. Ishimoto (281) 362-6554
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I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

**DECLARATION AND POWER OF ATTORNEY
FOR PATENT APPLICATION (continued)**

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Date

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Inventor's Signature

Date

SEQUENCE LISTING

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 Met Gln Lys Lys Gly Lys Tyr Ala Gln Leu Ile Gln Lys Met His Lys
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 Pro Lys Val Glu Ser Gln Ala Leu Ala Thr Ser Leu Glu Glu Ser Leu
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 Ala Gly Gly Tyr Met Val Ser Cys Ile Ile Phe Phe Phe Val Val Leu
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 Ile Val Phe Leu Thr Ile Phe Ser Phe Trp Trp Leu Ser Tyr Trp Leu
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 Leu Ser Leu Met Val Ile Ala Val Leu Leu Ile Val Ser Val Leu Ser
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<212> PRT
<213> homo sapiens

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35          40          45
Leu Arg Arg Lys Glu Arg Lys Leu Leu Glu Lys Cys Gly Leu Val Gln
50          55          60
Ser Leu Thr Ser Ile Thr Leu Phe Ile Ile Pro Thr Val Ala Thr Ala
65          70          75          80
Val Trp Val Leu Ile His Thr Ser Leu Lys Leu Lys Leu Thr Ala Ser
85          90          95
Met Ala Phe Ser Met Leu Ala Ser Leu Asn Leu Leu Arg Leu Ser Val
100          105          110
Phe Phe Val Pro Ile Ala Val Lys Gly Leu Thr Asn Ser Lys Ser Ala
115          120          125
Val Met Arg Phe Lys Lys Phe Phe Leu Gln Glu Ser Pro Val Phe Tyr
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Val Gln Thr Leu Gln Asp Pro Ser Lys Ala Leu Val Phe Glu Glu Ala
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Tyr	Val	Pro	Gln	Ala	Trp	Ile	Val	Ser	Gly	Asn	Ile	Arg	Glu	Asn	
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Glu	Ala	Thr	Ser	Asp	Met	Leu	Gln	Asp	Thr	Ala	Lys	Ile	Ala	Glu	Lys
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Ile	Val	Phe	Leu	Thr	Ile	Phe	Ser	Phe	Trp	Trp	Leu	Ser	Tyr	Trp	Leu
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Glu	Gln	Gly	Ser	Gly	Thr										

Glu Gln Leu Asp Gln Leu Leu Pro Ile Phe Ser Glu Gln Phe Leu Val
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 Pro Tyr Ile Leu Leu Met Gly Ala Ile Ile Met Val Ile Cys Phe Ile
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 Tyr Tyr Met Met Phe Lys Lys Ala Ile Gly Val Phe Lys Arg Leu Glu
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 Asn Tyr Ser Arg Ser Pro Leu Phe Ser His Ile Leu Asn Ser Leu Gln
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 Gln
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 <213> homo sapiens

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<213> homo sapiens

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Ile	Lys	Met	Tyr	Thr	Trp	Glu	Lys	Pro	Phe	Ala	Lys	Ile	Ile	Glu	Asp
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Phe	Phe	Val	Pro	Ile	Ala	Val	Lys	Gly	Leu	Thr	Asn	Ser	Lys	Ser	Ala
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Leu	Leu	Asp	Asp	Pro	Leu	Ser	Ala	Val	Asp	Ala	His	Val	Gly	Lys	His	
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Ile	Phe	Glu	Cys	Ile	Lys	Lys	Thr	Leu	Arg	Gly	Lys	Thr	Val	Val		
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Leu	Val	Thr	His	Gln	Leu	Gln	Tyr	Leu	Glu	Phe	Cys	Gly	Gln	Ile	Ile	
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Glu	Ala	Thr	Ser	Asp	Met	Leu	Gln	Asp	Thr	Ala	Lys	Ile	Ala	Glu	Lys	
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Pro	Lys	Val	Glu	Ser	Gln	Ala	Leu	Ala	Thr	Ser	Leu	Glu	Glu	Ser	Leu	
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Glu	Glu	Gly	Ser	Leu	Ser	Trp	Arg	Val	Tyr	His	His	Tyr	Ile	Gln	Ala	
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Ala	Gly	Gly	Tyr	Met	Val	Ser	Cys	Ile	Ile	Phe	Phe	Phe	Val	Val	Leu	
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Ile	Val	Phe	Leu	Thr	Ile	Phe	Ser	Phe	Trp	Trp	Leu	Ser	Tyr	Trp	Leu	
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Ser	Ser	Gly	Ile	Phe	Thr	Lys	Val	Thr	Arg	Lys	Ala	Ser	Thr	Ala	Leu	
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Asp	Thr	Ile	Pro	Ile	Gly	Arg	Leu	Asn	Cys	Phe	Ala	Gly	Asp	Leu		
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Glu	Gln	Leu	Asp	Gln	Leu	Leu	Pro	Ile	Phe	Ser	Glu	Gln	Phe	Leu	Val	
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Pro	Tyr	Ile	Leu	Leu	Met	Gly	Ala	Ile	Ile	Met	Val	Ile	Cys	Phe	Ile	
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Tyr	Tyr	Met	Met	Phe	Lys	Lys	Ala	Ile	Gly	Val	Phe	Lys	Arg	Leu	Glu	
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Asn	Tyr	Ser	Arg	Ser	Pro	Leu	Phe	Ser	His	Ile	Leu	Asn	Ser	Leu	Gln	
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 Leu Ser Ser Thr Arg Trp Met Ala Leu Arg Leu Glu Ile Met Thr Asn
 725 730 735
 Leu Val Thr Leu Ala Val Ala Leu Phe Val Ala Phe Gly Ile Ser Ser
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 Thr Pro Tyr Ser Phe Lys Val Met Ala Val Asn Ile Val Leu Gln Leu
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 Ala Ser Ser Phe Gln Ala Thr Ala Arg Ile Gly Leu Glu Thr Glu Ala
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 Gln Phe Thr Ala Val Glu Arg Ile Leu Gln Tyr Met Lys Met Cys Val
 785 790 795 800
 Ser Glu Ala Pro Leu His Met Glu Gly Thr Ser Cys Pro Gln Gly Trp
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 Pro Gln His Gly Glu Ile Ile Phe Gln Asp Tyr His Met Lys Tyr Arg
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 Asp Asn Thr Pro Thr Val Leu His Gly Ile Asn Leu Thr Ile Arg Gly
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 His Glu Val Val Gly Ile Val Gly Arg Thr Gly Ser Gly Lys Ser Ser
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 Leu Ile Asp Gly Val Asp Ile Cys Ser Ile Gly Leu Glu Asp Leu Arg
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 Ser Lys Leu Ser Val Ile Pro Gln Asp Pro Val Leu Leu Ser Gly Thr
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 Ile Arg Phe Asn Leu Asp Pro Phe Asp Arg His Thr Asp Gln Gln Ile
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 Trp Asp Ala Leu Glu Arg Thr Phe Leu Thr Lys Ala Ile Ser Lys Phe
 930 935 940
 Pro Lys Lys Leu His Thr Asp Val Val Glu Asn Gly Gly Asn Phe Ser
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 Val Gly Glu Arg Gln Leu Leu Cys Ile Ala Arg Ala Val Leu Arg Asn
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 Ser Lys Ile Ile Leu Ile Asp Glu Ala Thr Ala Ser Ile Asp Met Glu
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 995 1000 1005
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 His Ile Leu Val Met Gly Asn Gly Lys Val Val Glu Phe Asp Arg Pro
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<211> 1488

<212> DNA

<213> homo sapiens

<400> 7

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<210> 8

<211> 496

<212> PRT

<213> homo sapiens

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Ile	Lys	Met	Tyr	Thr	Trp	Glu	Lys	Pro	Phe	Ala	Lys	Ile	Ile	Glu	Asp
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Ser	Leu	Thr	Ser	Ile	Thr	Leu	Phe	Ile	Ile	Pro	Thr	Val	Ala	Thr	Ala
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Met	Ala	Phe	Ser	Met	Leu	Ala	Ser	Leu	Asn	Leu	Leu	Arg	Leu	Ser	Val
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Thr	Leu	Ser	Trp	Gln	Gln	Thr	Cys	Pro	Gly	Ile	Val	Asn	Gly	Ala	Leu
			165					170					175		
Glu	Leu	Glu	Arg	Asn	Gly	His	Ala	Ser	Glu	Gly	Met	Thr	Arg	Pro	Arg
			180					185				190			
Asp	Ala	Leu	Gly	Pro	Glu	Glu	Glu	Gly	Asn	Ser	Leu	Gly	Pro	Glu	Leu

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His Lys Ile Asn Leu Val Val Ser Lys Gly Met Met Leu Gly Val Cys		
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Gly Asn Thr Gly Ser Gly Lys Ser Ser Leu Leu Ser Ala Ile Leu Glu		
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Glu Met His Leu Leu Glu Gly Ser Val Gly Val Gln Gly Ser Leu Ala		
245	250	255
Tyr Val Pro Gln Gln Ala Trp Ile Val Ser Gly Asn Ile Arg Glu Asn		
260	265	270
Ile Leu Met Gly Gly Ala Tyr Asp Lys Ala Arg Tyr Leu Gln Val Leu		
275	280	285
His Cys Cys Ser Leu Asn Arg Asp Leu Glu Leu Leu Pro Phe Gly Asp		
290	295	300
Met Thr Glu Ile Gly Glu Arg Gly Leu Asn Leu Ser Gly Gly Gln Lys		
305	310	315
Gln Arg Ile Ser Leu Ala Arg Ala Val Tyr Ser Asp Arg Gln Ile Tyr		
325	330	335
Leu Leu Asp Asp Pro Leu Ser Ala Val Asp Ala His Val Gly Lys His		
340	345	350
Ile Phe Glu Glu Cys Ile Lys Lys Thr Leu Arg Gly Lys Thr Val Val		
355	360	365
Leu Val Thr His Gln Leu Gln Tyr Leu Glu Phe Cys Gly Gln Ile Ile		
370	375	380
Leu Leu Glu Asn Gly Lys Ile Cys Glu Asn Gly Thr His Ser Glu Leu		
385	390	395
Met Gln Lys Lys Gly Lys Tyr Ala Gln Leu Ile Gln Lys Met His Lys		
405	410	415
Glu Ala Thr Ser Val Phe Arg Cys Pro Met Ser Phe Phe Asp Thr Ile		
420	425	430
Pro Ile Gly Arg Leu Leu Asn Cys Phe Ala Gly Asp Leu Glu Gln Leu		
435	440	445
Asp Gln Leu Leu Pro Ile Phe Ser Glu Gln Phe Leu Val Leu Ser Leu		
450	455	460
Met Val Ile Ala Val Leu Leu Ile Val Ser Val Leu Ser Pro Tyr Ile		
465	470	475
Leu Leu Met Gly Ala Ile Ile Met Val Ile Cys Phe Ile Tyr Tyr Met		
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<210> 9
 <211> 1626
 <212> DNA
 <213> homo sapiens

<400> 9

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<210> 10
<211> 542
<212> PRT
<213> homo sapiens

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35 40 45
Leu Arg Arg Lys Glu Arg Lys Leu Leu Glu Lys Cys Gly Leu Val Gln
50 55 60
Ser Leu Thr Ser Ile Thr Leu Phe Ile Ile Pro Thr Val Ala Thr Ala
65 70 75 80
Val Trp Val Leu Ile His Thr Ser Leu Lys Leu Lys Leu Thr Ala Ser
85 90 95
Met Ala Phe Ser Met Leu Ala Ser Leu Asn Leu Leu Arg Leu Ser Val
100 105 110
Phe Phe Val Pro Ile Ala Val Lys Gly Leu Thr Asn Ser Lys Ser Ala
115 120 125
Val Met Arg Phe Lys Lys Phe Phe Leu Gln Glu Ser Pro Val Phe Tyr
130 135 140
Val Gln Thr Leu Gln Asp Pro Ser Lys Ala Leu Val Phe Glu Glu Ala
145 150 155 160
Thr Leu Ser Trp Gln Gln Thr Cys Pro Gly Ile Val Asn Gly Ala Leu
165 170 175
Glu Leu Glu Arg Asn Gly His Ala Ser Glu Gly Met Thr Arg Pro Arg
180 185 190
Asp Ala Leu Gly Pro Glu Glu Glu Gly Asn Ser Leu Gly Pro Glu Leu
195 200 205
His Lys Ile Asn Leu Val Val Ser Lys Gly Met Met Leu Gly Val Cys
210 215 220
Gly Asn Thr Gly Ser Gly Lys Ser Ser Leu Leu Ser Ala Ile Leu Glu
225 230 235 240
Glu Met His Leu Leu Glu Gly Ser Val Gly Val Gln Gly Ser Leu Ala
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Met	Thr	Glu	Ile	Gly	Glu	Arg	Gly	Leu	Asn	Leu	Ser	Gly	Gly	Gln	Lys
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Glu	Ala	Thr	Ser	Val	Phe	Arg	Cys	Pro	Met	Ser	Phe	Phe	Asp	Thr	Ile
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	450					455					460				
Met	Val	Ile	Ala	Val	Leu	Leu	Ile	Val	Ser	Val	Leu	Ser	Pro	Tyr	Ile
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Leu	Leu	Met	Gly	Ala	Ile	Ile	Met	Val	Ile	Cys	Phe	Ile	Tyr	Tyr	Met
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<212> DNA

<213> homo sapiens

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<210> 12

<211> 900

<212> PRT

<213> homo sapiens

<400> 12

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Ile Lys Met Tyr Thr Trp Glu Lys Pro Phe Ala Lys Ile Ile Glu Asp
 35          40          45
Leu Arg Arg Lys Glu Arg Lys Leu Leu Glu Lys Cys Gly Leu Val Gln
 50          55          60
Ser Leu Thr Ser Ile Thr Leu Phe Ile Ile Pro Thr Val Ala Thr Ala
 65          70          75          80
Val Trp Val Leu Ile His Thr Ser Leu Lys Leu Lys Leu Thr Ala Ser
 85          90          95
Met Ala Phe Ser Met Leu Ala Ser Leu Asn Leu Leu Arg Leu Ser Val
 100         105         110
Phe Phe Val Pro Ile Ala Val Lys Gly Leu Thr Asn Ser Lys Ser Ala
 115         120         125
Val Met Arg Phe Lys Lys Phe Phe Leu Gln Glu Ser Pro Val Phe Tyr
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His	Lys	Ile	Asn	Leu	Val	Val	Ser	Lys	Gly	Met	Met	Leu	Gly	Val	Cys	210	215	220	
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Glu	Met	His	Leu	Leu	Glu	Gly	Ser	Val	Gly	Val	Gln	Gly	Ser	Leu	Ala	245	250	255	
Tyr	Val	Pro	Gln	Gln	Ala	Trp	Ile	Val	Ser	Gly	Asn	Ile	Arg	Glu	Asn	260	265	270	
Ile	Leu	Met	Gly	Gly	Ala	Tyr	Asp	Lys	Ala	Arg	Tyr	Leu	Gln	Val	Leu	275	280	285	
His	Cys	Cys	Ser	Leu	Asn	Arg	Asp	Leu	Glu	Leu	Leu	Pro	Phe	Gly	Asp	290	295	300	
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Gln	Arg	Ile	Ser	Leu	Ala	Arg	Ala	Val	Tyr	Ser	Asp	Arg	Gln	Ile	Tyr	325	330	335	
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Ile	Phe	Glu	Glu	Cys	Ile	Lys	Lys	Thr	Leu	Arg	Gly	Lys	Thr	Val	Val	355	360	365	
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Glu	Ala	Thr	Ser	Val	Phe	Arg	Cys	Pro	Met	Ser	Phe	Phe	Asp	Thr	Ile	420	425	430	
Pro	Ile	Gly	Arg	Leu	Leu	Asn	Cys	Phe	Ala	Gly	Asp	Leu	Glu	Gln	Leu	435	440	445	
Asp	Gln	Leu	Leu	Pro	Ile	Phe	Ser	Glu	Gln	Phe	Leu	Val	Leu	Ser	Leu	450	455	460	
Met	Val	Ile	Ala	Val	Leu	Leu	Ile	Val	Ser	Val	Leu	Ser	Pro	Tyr	Ile	465	470	475	480
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Arg	Ser	Pro	Leu	Phe	Ser	His	Ile	Leu	Asn	Ser	Leu	Gln	Gly	Leu	Ser	515	520	525	
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Thr	Arg	Trp	Met	Ala	Leu	Arg	Leu	Glu	Ile	Met	Thr	Asn	Leu	Val	Thr	565	570	575	
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<212> PRT
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Tyr Lys Thr Tyr Thr Leu Gln Asp Gly Pro Trp Ser Gln Gln Glu Arg
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Asn Pro Glu Ala Pro Gly Arg Ala Ala Val Pro Pro Trp Gly Lys Tyr
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Cys	Tyr	Gly	Pro	Leu	Val	Leu	Ile	Thr	Cys	Ala	Ser	Leu	Val	Ile	Cys
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Ser	Ile	Ser	Ser	Tyr	Phe	Ile	Ile	Gly	Tyr	Thr	Ala	Phe	Ile	Ala	Ile
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Glu	Arg	Lys	Leu	Leu	Glu	Lys	Cys	Gly	Leu	Val	Gln	Ser	Leu	Thr	Ser
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Ile	Thr	Leu	Phe	Ile	Ile	Pro	Thr	Val	Ala	Thr	Ala	Val	Trp	Val	Leu
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Lys	Lys	Phe	Phe	Leu	Gln	Glu	Ser	Pro	Val	Phe	Tyr	Val	Gln	Thr	Leu
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Gln	Asp	Pro	Ser	Lys	Ala	Leu	Val	Phe	Glu	Glu	Ala	Thr	Leu	Ser	Trp
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Gly Lys Ile Cys Glu Asn Gly Thr His Ser Glu Leu Met Gln Lys Lys		
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Gly Lys Tyr Ala Gln Leu Ile Gln Lys Met His Lys Glu Ala Thr Ser		
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Ser Gln Ala Leu Ala Thr Ser Leu Glu Glu Ser Leu Asn Gly Asn Ala		
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Val Pro Glu His Gln Leu Thr Gln Glu Glu Glu Met Glu Glu Gly Ser		
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Leu Ser Trp Arg Val Tyr His His Tyr Ile Gln Ala Ala Gly Gly Tyr		
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Gln Arg Thr Ala Ile Arg Phe Gln Ala Ala Val Ser Ser Phe Ala Phe	
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Glu Lys Leu Ile Gln Phe Lys Ser Val Ile His Ile Thr Ser Gly Glu	
245 250 255	
Ala Ile Ser Phe Phe Thr Gly Asp Val Asn Tyr Leu Phe Glu Gly Val	
260 265 270	
Cys Tyr Gly Pro Leu Val Leu Ile Thr Cys Ala Ser Leu Val Ile Cys	
275 280 285	
Ser Ile Ser Ser Tyr Phe Ile Ile Gly Tyr Thr Ala Phe Ile Ala Ile	
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Leu Cys Tyr Leu Leu Val Phe Pro Leu Glu Val Phe Met Thr Arg Met	
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Ala Val Lys Ala Gln His His Thr Ser Glu Val Ser Asp Gln Arg Ile	
325 330 335	
Arg Val Thr Ser Glu Val Leu Thr Cys Ile Lys Leu Ile Lys Met Tyr	
340 345 350	
Thr Trp Glu Lys Pro Phe Ala Lys Ile Ile Glu Asp Leu Arg Arg Lys	


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<211> 1379

<212> PRT

<213> homo sapiens

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Asn Pro Glu Ala Pro Gly Arg Ala Ala Val Pro Pro Trp Gly Lys Tyr			
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Asp Ala Ala Leu Arg Thr Met Ile Pro Phe Arg Pro Lys Pro Arg Phe			
65	70	75	80
Pro Ala Pro Gln Pro Leu Gly Leu Phe Ser Tyr Leu Thr Val Ser Trp			
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Leu Thr Pro Leu Met Ile Gln Ser Leu Arg Ser Arg Leu Asp Glu Asn			
100	105	110	
Thr Ile Pro Pro Leu Ser Val His Asp Ala Ser Asp Lys Asn Val Gln			
115	120	125	
Arg Leu His Arg Leu Trp Glu Glu Val Ser Arg Arg Gly Ile Glu			
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Lys Ala Ser Val Leu Leu Val Met Leu Arg Phe Gln Arg Thr Arg Leu			
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Ile Phe Asp Ala Leu Leu Gly Ile Cys Phe Cys Ile Ala Ser Val Leu			
165	170	175	
Gly Pro Ile Leu Ile Ile Pro Lys Ile Leu Glu Tyr Ser Glu Glu Gln			
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Leu Gly Asn Val Val His Gly Val Gly Leu Cys Phe Ala Leu Phe Leu			
195	200	205	
Ser Glu Cys Val Lys Ser Leu Ser Phe Ser Ser Ser Trp Ile Ile Asn			
210	215	220	
Gln Arg Thr Ala Ile Arg Phe Gln Ala Ala Val Ser Ser Phe Ala Phe			
225	230	235	240
Glu Lys Leu Ile Gln Phe Lys Ser Val Ile His Ile Thr Ser Gly Glu			
245	250	255	
Ala Ile Ser Phe Phe Thr Gly Asp Val Asn Tyr Leu Phe Glu Gly Val			
260	265	270	
Cys Tyr Gly Pro Leu Val Leu Ile Thr Cys Ala Ser Leu Val Ile Cys			
275	280	285	
Ser Ile Ser Ser Tyr Phe Ile Ile Gly Tyr Thr Ala Phe Ile Ala Ile			
290	295	300	
Leu Cys Tyr Leu Leu Val Phe Pro Leu Glu Val Phe Met Thr Arg Met			
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Ala Val Lys Ala Gln His His Thr Ser Glu Val Ser Asp Gln Arg Ile			
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Arg Val Thr Ser Glu Val Leu Thr Cys Ile Lys Leu Ile Lys Met Tyr			
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Thr Trp Glu Lys Pro Phe Ala Lys Ile Ile Glu Asp Leu Arg Arg Lys			
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Lys Lys Phe Phe Leu Gln Glu Ser Pro Val Phe Tyr Val Gln Thr Leu			

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Ser Gly Lys Ser Ser	Leu Leu Ser Ala Ile	Leu Glu Glu Met His Leu		
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Leu Glu Gly Ser Val	Gly Val Gln Gly	Ser Leu Ala Tyr Val Pro Gln		
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Gln Ala Trp Ile Val	Ser Gly Asn Ile Arg	Glu Asn Ile Leu Met Gly		
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Gly Ala Tyr Asp Lys	Ala Arg Tyr Leu Gln	Val Leu His Cys Cys Ser		
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Leu Asn Arg Asp Leu	Glu Leu Leu Pro Phe	Gly Asp Met Thr Glu Ile		
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Gly Glu Arg Gly Leu	Asn Leu Ser Gly Gly	Gln Lys Gln Arg Ile Ser		
	625	630	635	640
Leu Ala Arg Ala Val	Tyr Ser Asp Arg Gln	Ile Tyr Leu Leu Asp Asp		
	645	650	655	
Pro Leu Ser Ala Val	Asp Ala His Val Gly	Lys His Ile Phe Glu Glu		
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Cys Ile Lys Lys Thr	Leu Arg Gly Lys Thr	Val Val Leu Val Thr His		
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Gln Leu Gln Tyr Leu	Glu Phe Cys Gly Gln	Ile Ile Leu Leu Glu Asn		
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Gly Lys Ile Cys Glu	Asn Gly Thr His Ser	Glu Leu Met Gln Lys Lys		
	705	710	715	720
Gly Lys Tyr Ala Gln	Leu Ile Gln Lys Met	His Lys Glu Ala Thr Ser		
	725	730	735	
Asp Met Leu Gln Asp	Thr Ala Lys Ile Ala	Glu Lys Pro Lys Val Glu		
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Ser Gln Ala Leu Ala	Thr Ser Leu Glu Glu	Ser Leu Asn Gly Asn Ala		
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Val Pro Glu His Gln	Leu Thr Gln Glu Glu	Glu Met Glu Glu Gly Ser		
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Leu Ser Trp Arg Val	Tyr His His Tyr Ile	Gln Ala Ala Gly Gly Tyr		
	785	790	795	800
Met Val Ser Cys Ile	Ile Phe Phe Phe Val	Val Leu Ile Val Phe Leu		
	805	810	815	
Thr Ile Phe Ser Phe	Trp Trp Leu Ser Tyr	Trp Leu Glu Gln Gly Ser		
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Gly Thr Asn Ser Ser	Arg Glu Ser Asn Gly	Thr Met Ala Asp Leu Gly		
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Asn Ile Ala Asp Asn	Pro Gln Leu Ser Phe	Tyr Gln Leu Val Tyr Gly		
	850	855	860	
Leu Asn Ala Leu Leu	Leu Ile Cys Val Gly	Val Cys Ser Ser Gly Ile		
	865	870	875	880
Phe Thr Lys Val Thr	Arg Lys Ala Ser Thr	Ala Leu His Asn Lys Leu		
	885	890	895	
Phe Asn Lys Val Phe	Arg Cys Pro Met Ser	Phe Phe Asp Thr Ile Pro		

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 Ser Leu Arg

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<212> PRT

<213> homo sapiens

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Asn Pro Glu Ala Pro Gly Arg Ala Ala Val Pro Pro Trp Gly Lys Tyr
 50          55          60
Asp Ala Ala Leu Arg Thr Met Ile Pro Phe Arg Pro Lys Pro Arg Phe
 65          70          75          80
Pro Ala Pro Gln Pro Leu Gly Leu Phe Ser Tyr Leu Thr Val Ser Trp
 85          90          95
Leu Thr Pro Leu Met Ile Gln Ser Leu Arg Ser Arg Leu Asp Glu Asn
100          105          110
Thr Ile Pro Pro Leu Ser Val His Asp Ala Ser Asp Lys Asn Val Gln
115          120          125
Arg Leu His Arg Leu Trp Glu Glu Glu Val Ser Arg Arg Gly Ile Glu
130          135          140
Lys Ala Ser Val Leu Leu Val Met Leu Arg Phe Gln Arg Thr Arg Leu
145          150          155          160
Ile Phe Asp Ala Leu Leu Gly Ile Cys Phe Cys Ile Ala Ser Val Leu
165          170          175
Gly Pro Ile Leu Ile Ile Pro Lys Ile Leu Glu Tyr Ser Glu Glu Gln
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Cys Tyr Gly Pro Leu Val Leu Ile Thr Cys Ala Ser Leu Val Ile Cys
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Ser Ile Ser Ser Tyr Phe Ile Ile Gly Tyr Thr Ala Phe Ile Ala Ile
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 <212> DNA
 <213> homo sapiens

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Gln Arg Thr Ala Ile Arg Phe Gln Ala Ala Val Ser Ser Phe Ala Phe
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275          280          285
Ser Ile Ser Ser Tyr Phe Ile Ile Gly Tyr Thr Ala Phe Ile Ala Ile
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